Original Article

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Promoter Methylation and Gene Expression in Human CD34$^+$ Stem Cells Derived Erythroid Lineage by MicroRNA

Mehdi Azad, PhD[1], Farshad Forooghi, PhD[2], Hassan Ehteram, PhD[3], Mousa Vatanmakan, Msc[4], Hajar Nasiri, Msc[5], Naser Mobarra, PhD[6,*]

1. Department of Medical Laboratory Sciences, Faculty of Allied Medicine, Qazvin University of Medical Sciences, Qazvin, Iran
2. Department of Immunology, Qazvin University of Medical Sciences, Qazvin, Iran
3. Department of Pathology, School of Medicine, Kashan University of Medical Sciences, Kashan, Iran
4. Department of Hematology, Faculty of Allied Medicine, Tehran University of Medical sciences, Tehran, Iran
5. Hematology-Oncology and Stem cell Transplantation Research Center, Tehran university of Medical Science, Tehran, Iran
6. Stem Cell Research Center, Department of Biochemistry, School of Medicine, Golestan University of Medical Sciences, Gorgan, Iran.

*Corresponding author: Naser Mobarra. PhD, Stem Cell Research Center, Department of Biochemistry, School of Medicine, Golestan University of Medical Sciences, Gorgan, Iran . E-mail: mobarra@goums.ac.ir.

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Abstract

Background: Stem Cell differentiation is a process composed of vast variety of factors which are controlled by a network of certain mechanisms. This study aims to determine the possible role of DNA methylation, a potent regulator of VHL, ECAD and RUNX3 genes during Erythroid differentiation driven by miR-451.

Materials and Methods: To determine the methylation status of promoters and the expression levels of VHL, ECAD and RUNX3 genes, Methylation Specific PCR (MSP) and real-time PCR were used, respectively, on both Cord Blood CD34+ Hematopoietic Stem Cells and differentiated cells. To measure the expression levels of mir-451, mirna qpcr technique was used.

Results: Our findings demonstrated a similar methylation pattern for the target genes before and after differentiation by miR-451. However, the expression levels were significantly increased after differentiation. Gene expression and surface marker analysis results further confirmed the potential of miR-451 for driving erythroid differentiation from hematopoietic stem cells.

Discussion: Our findings ruled out DNA methylation effect on the regulation of VHL, ECAD, and RUNX3 genes during miR-451 mediated erythroid differentiation. However, having CpG islands in their promoters, these three genes are candidates to be controlled by methylation which may not able to be detected by MSP method.

Conclusion: Taken together in this study we have shown a successful erythroid differentiation mediated by miR-451 which is at least in part, independent of DNA methylation. Further understanding of the underlying mechanisms driven by eryhtroid differentiation may lead to therapeutic measures to alter disorders of hematopoietic stem cell differentiation.

Keywords: DNA methylation, Erythroid differentiation, Gene expression, MiR-451, Stem cell

Introduction

Myeloid, lymphoid, and erythroid lineages are three different cell lineages of hematopoietic system (1, 2). Differentiation into these three functional distinct lineages from hematopoietic stem cells (HSCs) includes myriad of molecular interactions that are able to regulate HSC self-renewal and lineage fate (1, 3-5). Identification of specific molecular pathways which regulate HSC differentiation, self-renewal, and proliferation remains a fundamental aim of either basic or clinical biology (5, 6). Proper lineage specification can be prevented by different genetic and epigenetic aberrations affecting differentiation and/or proliferation of HSCs and ultimately lead to severe diseases like myelodysplasia and leukemia